

ESTIMATION OF AN UPPER TOLERANCE LIMIT FOR SMALL-SAMPLES
CONTAINING OBSERVATIONS BELOW THE LIMIT OF QUANTITATION

by

DONGLIN YAN

B.S, Northern Arizona University, 2012

A REPORT

submitted in partial fulfillment of the requirements for the degree

MASTER OF SCIENCE

Department of Statistics
College of Art and Science

KANSAS STATE UNIVERSITY
Manhattan, Kansas

2014

Approved by:

Major Professor
Christopher I. Vahl

Abstract

Chemicals and drugs applied to animals used in meat production often have the potential to cause adverse effects on human consumers. To ensure safety, a withdrawal period, i.e. the minimum time allowed between application of the drug and entry of the animal into the food supply, must be determined for each drug used on food-producing animals. The withdrawal period is based on an upper tolerance limit at a given time point. It is not uncommon that the concentration of the drug in some tissue samples to be measured at a level below the limit of quantitation (LOQ). Because the measurement of the tissue concentration cannot be confidently determined with enough precision, these types of observations are often treated as if they were left censored where the censoring value is equal to the limit of quantitation. Several methods are commonly used in practice to deal with this situation. The simplest methods are either to exclude observations below the limit of quantitation or to replace those values with zero, LOQ or $\frac{1}{2}$ LOQ. Previous studies have shown that these methods result in biased estimation of the population mean and population variance. Alternatively, one could incorporate censoring into the likelihood and compute the maximum likelihood estimate (MLE) for the population mean and variance assuming a normal or lognormal distribution. These estimates are also biased but it has been shown that they are asymptotically unbiased. However, it is not clear yet how these various methods affect estimation of the upper tolerance limit, especially when the sample size is small, e.g. less than 35. In this report, we will examine the effects of substituting the LOQ or $\frac{1}{2}$ LOQ for censored values as well as using the MLEs of the mean and variance in the construction of an upper tolerance limit for a normal population through simulation. Additionally, we propose a modified substitution method where observations below the LOQ are replaced by functions of the order statistics of non-censored observations under an assumption of symmetry. Its performance relative to the above methods will also be evaluated in the simulation study. In the end, the results from this study will be applied to an environmental study.

Table of Contents

List of Figures	iv
List of Tables	v
Acknowledgements	vi
Chapter 1 - Introduction	1
1.1 Background and the Problem	1
1.2 Concepts and Terminologies	3
1.2.1 Limit of Blank, Limit of Detection, Limit of Blank and Left Censoring	3
1.2.4 Tolerance Limit	5
Chapter 2 - Common Methods in Analysis of Left-Censored Data	7
2.1 Maximum Likelihood Estimation Method	7
2.2 Traditional Substitution Methods	8
2.3 The Modified Substitution Method	9
2.4 Example of the Candidate Methods	11
Chapter 3 - Simulation Design	12
Chapter 4 - Simulation Results and Conclusions	13
4.1 Coverage Rate of the Estimated Tolerance Limits	13
4.2 Evaluation of MLE Method	15
4.3 Evaluation of Traditional Substitution Methods	17
4.4 Evaluation of Modified Substitution Method	19
4.5 Conclusions	20
Chapter 5 - Case Study and Discussion	21
Bibliography	24
Appendix A - Summary of Simulation Results	25
The following tables summarized the simulation results. Recall that we have run the simulation 2500 rounds for each combination of percent of censoring and sample size. The hypothetical population from which random samples are drawn is $N(12, 3)$	
Appendix B - R code For Figure 4.2-4.5	30
Appendix C - Simulation SAS code	38

List of Figures

Figure 1.1 Graphical representation of the differences between LOB, LOD and LOQ	5
Figure 4.1 Coverage-rate of (0.9, 0.95) UTLs estimated by the candidate methods in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$).	14
Figure 4.2 Distribution of UTL estimated by MLE method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.	16
Figure 4.3 Distribution of UTL estimated by $\frac{1}{2}$ LOQ substitution method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.	18
Figure 4.4 Distribution of UTL estimated by LOQ substitution method in different scenarios (percent censoring = 10% ,20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.	18
Figure 5.1 Distribution of atrazine and log(Atrazine) for data in Table 5.1	22

List of Tables

Table 1.1 Hypothetical drug concentrations in liver and muscle tissue for 10 tested animals where LOQ=1.5 µg/k.	3
Table 2.1 Hypothetical muscle data after application of the ½ LOQ substitution, LOQ substitution, and modified substitution method, where LOQ=1.5 µg/k	10
Table 2.2 Sample mean \bar{x} , sample standard deviation s , estimated 90 th percentile and UTL calculated by MLE, MS, ½ LOQ and LOQ to the hypothetical data. LoQ=1.5 µg/k and MRL= 4.0 µg /kg	11
Table A.1 Mean of \bar{x} , estimated bias of \bar{x} , mean s , estimated bias of s , mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 10$	25
Table A.2 Mean of \bar{x} , estimated bias of \bar{x} , mean s , estimated bias of s , mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 20$	26
Table A.3 Mean of \bar{x} , estimated bias of \bar{x} , mean s , estimated bias of s , mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 30$	27
Table A.4 Mean of \bar{x} , estimated bias of \bar{x} , mean s , estimated bias of s , mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 45$	28
Table A.5 Mean of \bar{x} , estimated bias of \bar{x} , mean s , estimated bias of s , mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 60$	29

Acknowledgements

I cannot express enough thanks to my committee for their contribution, support and encouragement: Dr. Christopher Vahl, my committee chair; Dr. Leigh Murray; Dr. Abigail Jager; I sincerely offer my appreciation for the learning opportunities provided by my committee.

Thanks to the Department of Statistics, Kansas State University for providing me the opportunity to complete my Master's study. In the past two years, I learned a lot from the dedicated faculty, and had a wonderful time with my fellow classmates.

Chapter 1 - Introduction

1.1 Background and the Problem

Different types of chemicals or drugs are applied to animals used in meat production in order to treat or prevent diseases or to increase productivity. After applying a drug to an animal, the concentration of the active ingredient gradually decreases until it is depleted. Residues of pharmaceuticals in edible animal tissues can have adverse health effects on humans. Maximum Residue Limits (MRLs) for pesticide residues and residues of veterinary drugs are the maximum concentrations of residues to be permitted in or on a food, by national or regional legislation, according to Food and Agriculture Organization of the United Nations and World Health Organization (2009). Although MRLs may be different for different countries, they are generally determined for food producing animals by the maximum amount of veterinary drug that humans can consume daily without the risk of causing health problems. Hence, a sufficiently long withdrawal period is necessary after application of drugs to animals so that the meats on market have residue concentrations below the MRLs. On the other hand, a shorter withdrawal period is desired for economic reasons. For example, an animal producer may not want to use a drug with a long withdrawal period because this may cause delays in shipping treated animals to market, or in the case of dairy cows, force the producer to discard milk until the withdrawal period expires.

Both the US Food and Drug Association (FDA) and European Medicines Agency (EMA) have standardized methods to estimate the withdrawal period when residues of certain chemicals in all animal tissues have depleted to below a safe level for most of the animals. Standardized methods take samples of animals, measure their residue concentration amount in tissues at intervals of time and then use statistical approaches to model concentration amount over time. The withdrawal period is determined relative to the first time point where the upper tolerance limit for the 99th percentile for concentrations in a particular tissue group is below the predetermined MRL. (EMA, 1997)

However, as the concentration lowers, the issue of left censoring often arises. When measurements of a certain chemical residue are lower than the Limit of Quantification (LOQ), which will be formally defined later, the measured values are believed to be unreliable and hence, are reported as non-quantified rather than as the actual measured value. There are several

common ways to deal with observations below the LOQ. Among these are to set these values to a single constant equal to zero, one half of the LOQ, or the LOQ itself; discard these data points; or use the maximum likelihood method modified to handle left-censored observations. This report, by simulation, shows that the above ways to deal with non-quantified data will bias the estimation of the upper tolerance limit which will consequently bias the estimation of withdrawal period.

In this study, we focus on the estimation of the upper tolerance limit at one time point using data observed at only that single time point and thus do not consider the more complicated situation where concentrations decrease and are measured over time. This scenario does occur in practice and is not merely an oversimplification. For example, suppose that a new drug for dairy cows is being tested and will be given to the animals so that the concentrations of the drug in the animals' blood plasma and tissues will reach a steady state, i.e. concentrations of the drug in each cow will reach a maximum level and stay there, plus or minus some small error, over an extended period of time. The company developing the new drug will then want to demonstrate that the concentrations of the drug in edible tissues of cows treated with the drug at steady state are below the maximum residue limit (MRL) for each group of edible tissues. Let us suppose that tissues samples were taken from 10 cows at steady state. Samples of several types of tissue such as liver, kidney, heart, muscle and fat are usually harvested from each animal and the concentration of the drug in each tissue sample is measured in μg of the drug per kg of tissue. Because of their biological functions, some tissues such as liver and kidney tend to accumulate higher concentrations of the drug than other tissues such as fat and muscle. Therefore, a governmental agency will typically set a separate MRL for each tissue type. Suppose the MRLs for this new drug have been set to 40.0 and 4.0 $\mu\text{g}/\text{kg}$ for liver and muscle respectively, and for the 10 cows we observed, concentrations for liver and muscle given in the table below (Table 1.1). Let us further suppose that during development of the analytical method used to extract the drug from tissue in order to measure concentrations, the LOQ of the method was determined to be 1.5 $\mu\text{g}/\text{kg}$. Note that for the hypothetical liver measurements, all observations are above the LOQ, but there are two observations for muscle that have been reported as " < 1.5 ". In other words, these two observations are below the LOQ and may be considered to be left censored. Our now goal is to determine whether or not most members in the population from which the data was

sampled have concentrations of the drug lower than a given MRL. To do this we will compute an upper tolerance limit that takes into account the values observed below the LOQ.

Table 1.1 Hypothetical drug concentrations in liver and muscle tissue for 10 tested animals where LOQ=1.5 µg/k.

Cow ID	Liver	Muscle
1	3.0	2.7
2	23.0	<1.5
3	18.0	2.5
4	12.0	1.6
5	13.0	1.8
6	9.0	<1.5
7	27.5	2.2
8	14.0	2.7
9	21.0	1.6
10	7.5	3.7

1.2 Concepts and Terminologies

1.2.1 Limit of Blank, Limit of Detection, Limit of Blank and Left Censoring

As the concentration of chemicals gets lower, the measurements of the concentration value become less reliable. Differences between the true concentration values and the measured values are known as measurement error. There are two possible sources of measurement error: the bias and the random error. Let c be the real concentration of the chemical of interest. Suppose M_c is the measured concentration value given the true concentration is c , m_c is the observed value of M_c . Then we have $E(M_c) = c + f(c)$, where $f(c)$ is the bias of the measurement technique at concentration c . Also, suppose $\sigma_c = \sqrt{\text{Var}(M_c)}$ is the standard deviation among repeated measurements of samples with the same concentration value c . We can assume σ_c is also a function of c . Therefore, total error = $m_c - c = (E(M_c) + e) - c = f(c) + e$, where e is the random error. Usually, when the measured concentration m is below a predetermined threshold the measured value can no longer be trusted since the total error would be intolerably large. In this case, “<LOQ” or “N/Q” will be recorded into the data rather than the measured value.

There are several types of limits associated with the difficulty of measuring very small concentrations. Among these are limit of blank (LOB), limit of detection (LOD) and limit of quantitation. There are other definitions for each limit, but in this report, we use the definitions proposed by Clinical and Laboratory Standards Institute. The approved guideline EP17 (CLSI 2004), Protocols for Determination of Limits of Detection and Limits of Quantitation, provides a standard method for determining LOB, LOQ and LOQ.

The limit of blank, or LOB, is defined as the highest apparent chemical concentration expected to be found when replicates of a sample containing no chemicals are tested. LOB is estimated by measuring replicates of a blank sample and calculating the mean results and the standard deviation. Assuming measurements of blank samples are normally distributed with mean μ_B and standard deviation σ_B , denoted here as $N(\mu_B, \sigma_B)$, then letting $LOB = \mu_B + 1.645\sigma_B$ ensures $Pr(c=0 | M > LOB) < 0.05$. Therefore, if a measurement is greater than LOB, we will be at least 95% confident that the analyte is present. A 95 % confidence limit for LOB is estimated by $\bar{x}_B + 1.645 * SD_B$, where \bar{x}_B and SD_B are the sample mean and sample standard deviation of the measurements.

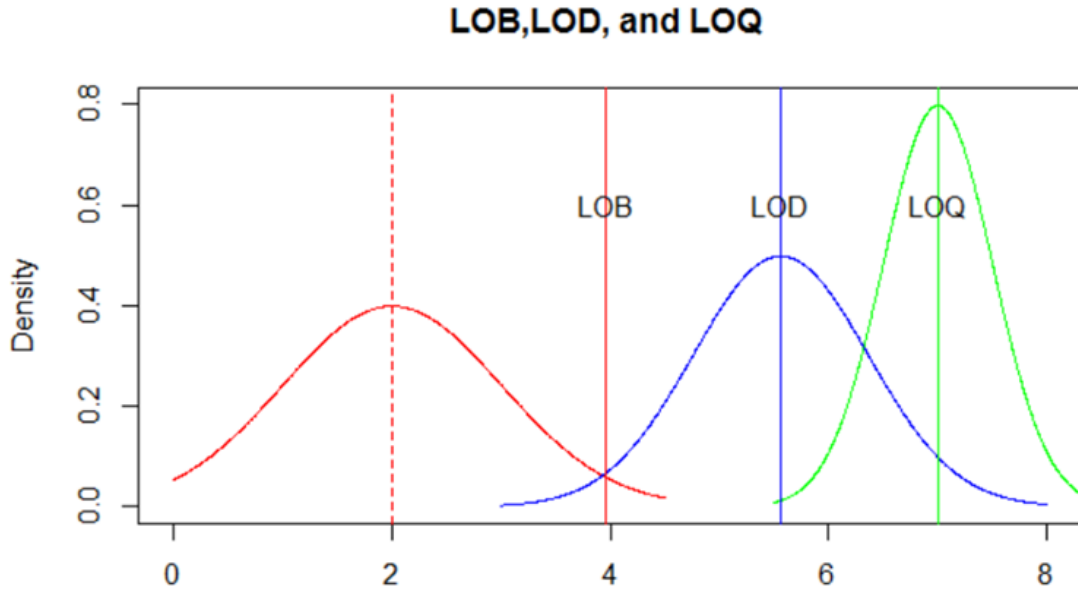
The limit of detection, or LOD, is the lowest analyte concentration likely to be reliably distinguished from the LOB and at which detection is feasible. It is estimated by repeatedly measuring samples with a very low but known concentration. Let SD_{low} denotes the standard deviation of measurements of the low concentration sample. A 95% confidence for LOQ is calculated as $LOD = LOB + (1.645 * SD_{low})$, which ensures $Pr(M > LOB | C = LOD) = 0.05$

The limit of quantitation, or LOQ, is, according to EP17, the lowest amount of analyte in a sample that can be quantitatively determined with stated acceptable precision and trueness, under stated experimental conditions. It is not only the lowest actual amount of analyte that can be reliably detected, but also at which the total measurement error (i.e. bias + random error) meets the laboratory's requirements for accuracy. The maximum amounts of measurement error that can be tolerated are determined before the estimation of LOQ. Let c be the real concentration of some analyte and M be the measurement. Assuming that the accuracy of a measurement increases as the concentration increases, $c \sim N(\mu, \sigma)$, and $M \sim N(c, \sigma_c)$, then we have the definition $LOQ = \min\{c : |f(c) + 2\sigma_c| < k\}$, where k is the predetermined amount of measurement error that can be tolerated. When $f(c) = 0$, the definition of LOQ can be simplified

to $LOQ = \min\{c : CV < p\}$, where $CV = \sigma_c / \mu$ is the coefficient of variance of the measurement and a typical value of p is 0.2.

Left censored data occurs when data points are known to be below a certain value but their exact values are unknown (Lambert et al., 1991). In this report, we assume that all censored observations have true values less than or equal to LOQ and there is only one limit of quantitation in the data.

Figure 1.1 Graphical representation of the differences between LOB, LOD and LOQ



1.2.4 Tolerance Limit

For a univariate population, a tolerance interval is an interval based on a random sample which is expected to contain a specified proportion (or more) of the sampled population (Krishnamoorthy and Mathew, 2009). Let α and p be predetermined constants between 0 and 1. Then a $(p, 1-\alpha)$ tolerance interval is expected to contain at least $p*100\%$ of the population at $1-\alpha$ confidence level. Here, the values $1-\alpha$ and p are said to be the confidence level and content of the tolerance interval respectively.

Similarly, a one-sided $(p, 1-\alpha)$ upper tolerance limit (UTL) is expected to be greater than at least $p*100\%$ of the population at a $1-\alpha$ confidence level. It can be shown that a $(p, 1-\alpha)$ UTL is simply an upper $1-\alpha$ confidence limit for the $100*p$ percentile of the population (Krishnamoorthy & Lian, 2011).

Suppose X is a random variable from some distribution with cumulative distribution function $F(x)$ and $\underline{x} = (x_1, x_2, \dots, x_n)$ is a random sample from the distribution. Let q_p denote the p^{th} percentile of the distribution. The $(p, 1-\alpha)$ tolerance limit is $U(\underline{x})$ such that $Pr(Pr(X \leq U(\underline{x}) | \underline{x}) \geq p) = 1-\alpha$.

Assuming $X \sim N(\mu, \sigma)$, a $(p, 1-\alpha)$ upper tolerance limit is

$$\bar{x} + k_1 s, \text{ where } k_1 = \frac{1}{\sqrt{n}} t_{n-1, 1-\alpha, Z_p \sqrt{n}}$$

The term k_1 is called a tolerance factor. The terms $n-1$, $1-\alpha$, and $Z_p \sqrt{n}$ are the degree of freedom, lower-tail proportion and non-centrality parameter of a non-central t distribution, respectively. Z_p represents the $100p$ percentile from a standard normal distribution. With respect to the dairy cow example in section 1.1, suppose we observe the sample average concentration \bar{x} and standard deviation s with $n=10$. We can be 95% confident that 90% of the dairy cow population at steady state has a concentration below the (0.90, 0.95) UTL, where the UTL is calculated as

$$\bar{x} + \frac{1}{\sqrt{10}} t_{9, 0.95, (Z_{0.90} \sqrt{10})} s = \bar{x} + 2.3209s$$

The estimation of UTL is a key component in determination of the withdrawal period. Unfortunately, the procedure of estimating the tolerance limit is often hindered by the observation of censored data. In the next chapter, some commonly used methods and a proposed new method for addressing the censored data issue are introduced. Similar studies regarding these types of methods usually focus on their performance in estimating the population mean and population variance. This report emphasizes the performance of these methods with respect to estimating the UTL when sample sizes are small.

Chapter 2 - Common Methods in Analysis of Left-Censored Data

2.1 Maximum Likelihood Estimation Method

The MLE method has widely been accepted in the analysis of censored data provided that the assumption(s) for the population distribution is plausible. The likelihood function of an estimator based on censored data looks like the usual likelihood function, but with less precise information given by the censored data. When none of the data has been censored, we can estimate the parameters μ and σ for a normal population by maximizing the likelihood function

$$L(\mu, \sigma) = \prod_{i=1}^n f(x_i), \text{ where } f(x_i) = \frac{1}{\sigma\sqrt{2\pi}} e^{-(x_i - \mu)^2 / 2\sigma^2}$$

The maximum likelihood estimators of μ and σ^2 are respectively $\hat{\mu} = \bar{x}$ and $\hat{\sigma}^2 = (n-1)s^2 / n$ which maximize the likelihood function $L(\mu, \sigma)$. With left censored data, it is assumed that each censored x_i is smaller than the LOQ, so the likelihood function can be revised to

$$L(\mu, \sigma) = \prod_{i=1}^n [f(x_i)]^{c_i} [F(LOQ)]^{1-c_i}$$

where $F(\cdot)$ is the cumulative distribution function of the distribution and c_i is a constant where $c_i = 0$ if $x_i \leq LOQ$ and $c_i = 1$ if $x_i > LOQ$ (Wolynetz, 1979).

It has been shown that for any MLE estimator $\hat{\theta}$, the MLE estimator of $f(\theta)$ is $f(\hat{\theta})$ for any real function $f(\cdot)$ (Casella & Berger, 2002). Therefore, the MLE of the p^{th} percentile is $\hat{P}_p = \bar{x} + Z_p s$, where Z_p is the p^{th} percentile of standard normal distribution, i.e. $N(0,1)$.

The upper confidence limit (UCL) can be calculated using the asymptotic properties of MLE estimators; $UCL = \hat{\theta} + Z_{1-\alpha} SE(\hat{\theta})$ where $SE(\hat{\theta})$ is the estimated standard error of $\hat{\theta}$. Since a $(p, 1-\alpha)$ UTL is equivalent to the $1-\alpha$ upper confidence limit for the $100p$ percentile of the population, we have

$$UTL = UCL = \hat{P}_p + Z_{1-\alpha} SE(\hat{P}_p).$$

2.2 Traditional Substitution Methods

The FDA recommends excluding data observed below the limit of detection from the calculation. EMEA-CVMP suggests setting the data below the limit of detection or quantitation to one-half of the respective limit (EMEA 1995). Setting the data to be equal to the quantitation limit is also a commonly used method in industry. Intuitively, setting censored data to its upper limit will make the analysis conservative and over-estimate the tolerance limit. However, the simulation results in this study suggest quite the opposite. Previous simulation studies have shown that these substitution methods are biased when estimating population the mean and population variance. (Hewett and Ganser, 2007) However, it appears that no studies have been conducted to evaluate these methods with respect to estimation of a tolerance limit. In this study, we will consider two widely used substitution methods: replacing the censored data with either one half of LOQ or the LOQ itself.

Referring to the dairy cow data in Table 1.1, Table 2.1 shows how traditional substitution methods replace the censored observations. The $\frac{1}{2}$ LOQ method uses data in column 3 to calculate UTL; the LOQ method uses the 4th column.

Seemingly, by substituting censored data by the LOQ looks like a more conservative method (over estimating tolerance limit), since the LOQ is larger than the censored data. Similarly, the $\frac{1}{2}$ LOQ method would be referred to as liberal.

2.3 The Modified Substitution Method

In this report, we propose and evaluate a modified version of the substitution methods. Since the normal distribution is symmetric about its mean and median, we can use information observed above the sample median in the upper tail of the sample (i.e. observations that are not censored) to replace the censored observations in the lower tail of the sample with values more meaningful than merely replacing them with LOQ or $\frac{1}{2}$ LOQ. For the case where less than 50% of the observations are left censored and the observations in the data are independently and identically sampled from $N(\mu, \sigma)$, let $x_{(i)}$ denote the i^{th} order statistic and $x_{(m)}$ denote the median where $i = 1, 2, \dots, n$. Create a new substitution data set by using modified order statistics defined as

$$x_{(i)}^* = \begin{cases} x_{(i)}, & \text{if } x_{(i)} > LOQ \\ 2x_{(m)} - x_{(n+1-i)}, & \text{if } x_{(i)} \leq LOQ \end{cases}$$

and then calculate the tolerance limit using the modified data $x_{(i)}^*, i = 1, 2, \dots, n$.

Suppose there are k censored data values in a data set of size n and $2k < n$. Then we can calculate $x_{(i)}^*$ for $i=1 \dots n$. After we substitute the censored observations, we can calculate

$$\bar{x}^* = \hat{\mu} = \frac{\sum_{i=1}^n x_{(i)}^*}{n}, \quad s^* = \hat{\sigma} = \frac{\sum_{i=1}^n (x_{(i)}^* - \bar{x}^*)^2}{n-1}, \quad \text{UTL} = \bar{x}^* + k_1 s^*$$

as estimators of μ , σ , and UTL where UTL is computed using the usual normal theory assumptions given in Section 1.2.4.

Referring to the dairy cows' data in **Table 1.1**, the median for muscle concentration is 2.0. Replace the first censored observation by $2 * 2 - 3.7 = 0.3$ and replace the second censored observation by $2 * 2 - 2.7 = 1.3$. Then the UTL is calculated using the 5th column in Table 2.1.

Table 2.1 Hypothetical muscle data after application of the $\frac{1}{2}$ LOQ substitution, LOQ substitution, and modified substitution method, where LOQ=1.5 $\mu\text{g}/\text{k}$

Cow ID	Muscle	$\frac{1}{2}\text{LOQ}$	LOQ	Modified Substitution
1	2.7	2.7	2.7	2.7
2	<1.5	0.75	1.5	1.3
3	2.5	2.5	2.5	2.5
4	1.6	1.6	1.6	1.6
5	1.8	1.8	1.8	1.8
6	<1.5	0.75	1.5	0.3
7	2.2	2.2	2.2	2.2
8	2.7	2.7	2.7	2.7
9	1.6	1.6	1.6	1.6
10	3.7	3.7	3.7	3.7

2.4 Example of the Candidate Methods

We will use the dairy cow study introduced in Chapter 1 as a numerical example to illustrate the four methods introduced above. With two censored observations in the muscle data, we will need to apply the described methods to those observations. The sample mean, standard deviation, 90 percentile and (90, 95) UTL were calculated for each method and are summarized in Table 2.2. Notice that the four methods give different estimations of the UTL. According to the MLE and LOQ method, we are 95% confident that at least 90% of cows have drug residue levels less than the MRL=4.0 µg/kg. On the contrary, the MS and ½ LOQ method suggest that there is no sufficient evidence to draw the same conclusion. As to which method we should trust, we will base our recommendation on the results of the simulation study introduced in Chapter 3.

Table 2.2 Sample mean \bar{x} , sample standard deviation s , estimated 90th percentile and UTL calculated by MLE, MS, ½ LOQ and LOQ to the hypothetical data. LoQ=1.5 µg/k and MRL= 4.0 µg /kg

	MLE	MS	½ LOQ	LOQ
\bar{x}	2.08	2.04	2.03	2.18
s	0.82	0.94	0.92	0.77
\hat{p}_{90}	3.13	3.25	3.21	3.17
UTL	3.72	4.21	4.17	3.97

Chapter 3 - Simulation Design

In this report, we assume the population distribution is normal. Without loss of generality, we assume the population mean is 12 and the standard deviation is 3. Percent of censored data and sample size are two important factors that have an influence on the performance of each analysis method; therefore we let these two factors vary. Let P stand for the percent of censored data. Because we will set the LOQ to a constant, the actual observed value of P will be a random variable. Here we set $E(P) = 10\%, 20\%, 30\%, 40\%$. The sample size within each round of simulation is set to $n = 10, 20, 30, 45, \text{ or } 60$. Let N be the number of replicates for each combination of $E(P)$ and n . The response variables of interest are the coverage rate of Tolerance Limit (denoted by \hat{p}) and average deviation of the UTL from the true population percentile (estimated bias, denoted by \hat{B}). In order to make the standard error of \hat{p} less than 0.01, we choose $N = 2500$.

Random samples of size n were drawn from the hypothesized $N(12, 3)$ population. We are assuming that P percent of the population is censored, so we let $\text{LOQ} =$ the P percentile of the population distribution. For $E(P) = 10\%, 20\%, 30\%, 40\%$, we have corresponding values of $\text{LOQ} = 8.1553, 9.4751, 10.4268, 11.2396$. Sample observations that are smaller than or equal to LOQ are marked as left censored.

Then we apply each candidate analysis method to each simulated dataset and repeat this procedure 2500 times for each combination of n and $E(P)$. We used the LIFEREG procedure in SAS[®] 9.3 to perform the MLE method; the SAS DATA Step and UNIVARIATE procedure were used to implement the traditional and modified substitution procedures. See appendix A for SAS code used in the simulation. For each repetition, we recorded the estimated mean the standard deviation, 90th percentile, and the 95% confidence level UTL of each candidate method. In addition to the candidate methods introduced above, we included a “control method” where we do not censor any observations and use the MLE method to estimate UTL. The purpose of this control method is to provide a benchmark for the candidate methods.

Chapter 4 - Simulation Results and Conclusions

We evaluate the performance of candidate methods from two perspectives. First, we evaluate the coverage rate of each candidate method. This is done by comparing the estimated UTL using different methods to 15.8447, which is the true 90th percentile of the hypothesized population. Since we are estimating a 90th percentile, 95% confidence level Tolerance Limit, the best candidate method should give a coverage rate closer to 0.95 than the other methods. Second, we will evaluate the bias and standard deviation of the estimated TLs. In order to estimate bias, we calculate the true Upper Tolerance Limit by $UTL = \mu + k_1 \sigma$, where $k_1 = \frac{1}{\sqrt{n}} t_{n-1, 1-\alpha(Z_p \sqrt{n})}$

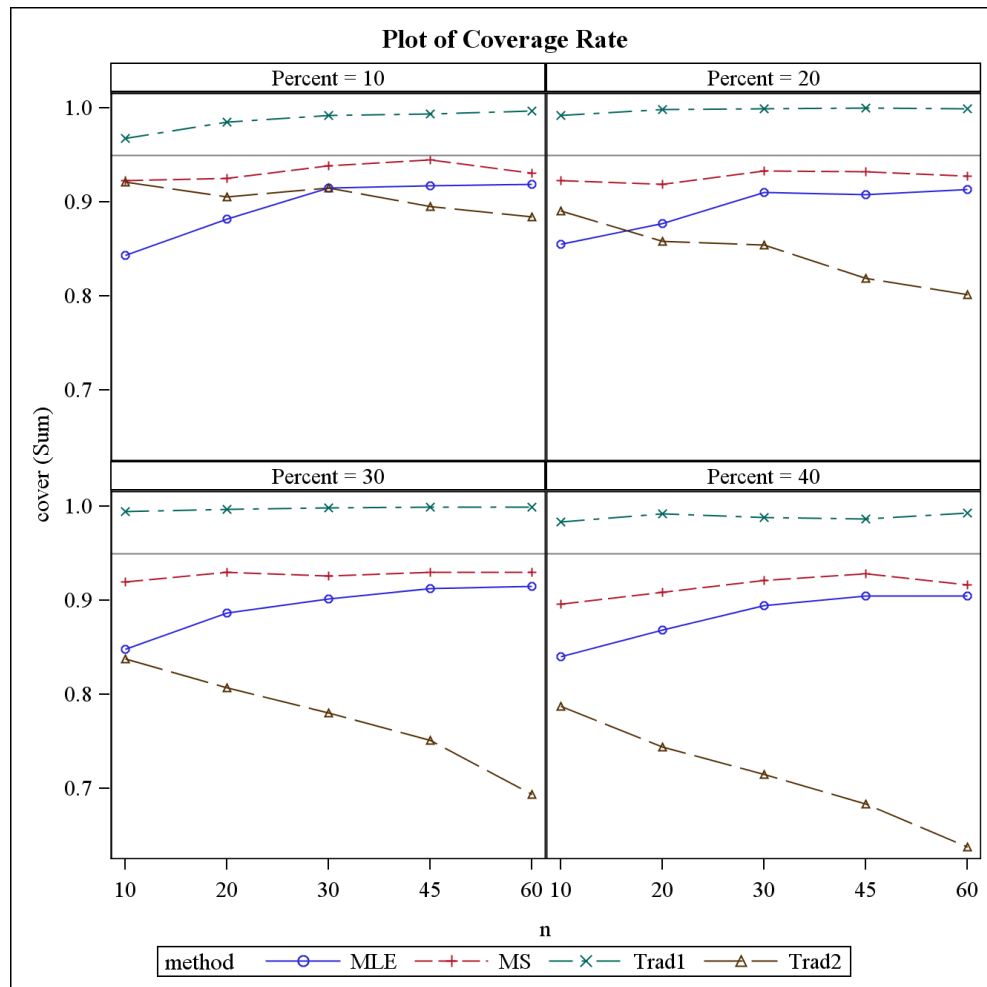
4.1 Coverage Rate of the Estimated Tolerance Limits

Coverage rate is an important feature of an UTL. If repeated samples were taken, and a $(p, 1-\alpha)$ UTL computed for each sample, $1-\alpha$ of the UTLs should be greater than or equal to the p^{th} percentile of the population. Since the data is censored, the coverage rate of UTLs will differ from $1-\alpha$. Coverage rates of different analysis methods are summarized in Figure 4.1. Cover rate of the MLE method approaches 0.95 as sample sizes increases, but it is too low to provide accurate estimation when sample size is small. The traditional substitution method 1 (brown) which replaces censored data by $\frac{1}{2}$ LOQ is not underestimating the UTL as previously expected. Even though it is replacing censored data by a smaller value, we are still over estimating the UTL. Also, the Traditional substitution method 2 (purple) which replaces censored data by LOQ is underestimating the UTL. These counter-intuitive results are caused by the biased estimation of σ , which led to the bias in estimating the Upper Tolerance Limit.

Coverage of the Modified Substitution method is outperforming the other methods, especially when sample sizes are small. Although it's not reaching 95% cover rate, it is doing much better than the MLE method for small samples.

Also notice that percent of censoring does not have too much influence on coverage rate for the MLE method and MS method. In practice, we do not know exactly what proportion of the population is censored, but based on this simulation, the MS method tends to give more reliable estimation at a given confidence level.

Figure 4.1 Coverage-rate of (0.9, 0.95) UTLs estimated by the candidate methods in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$).

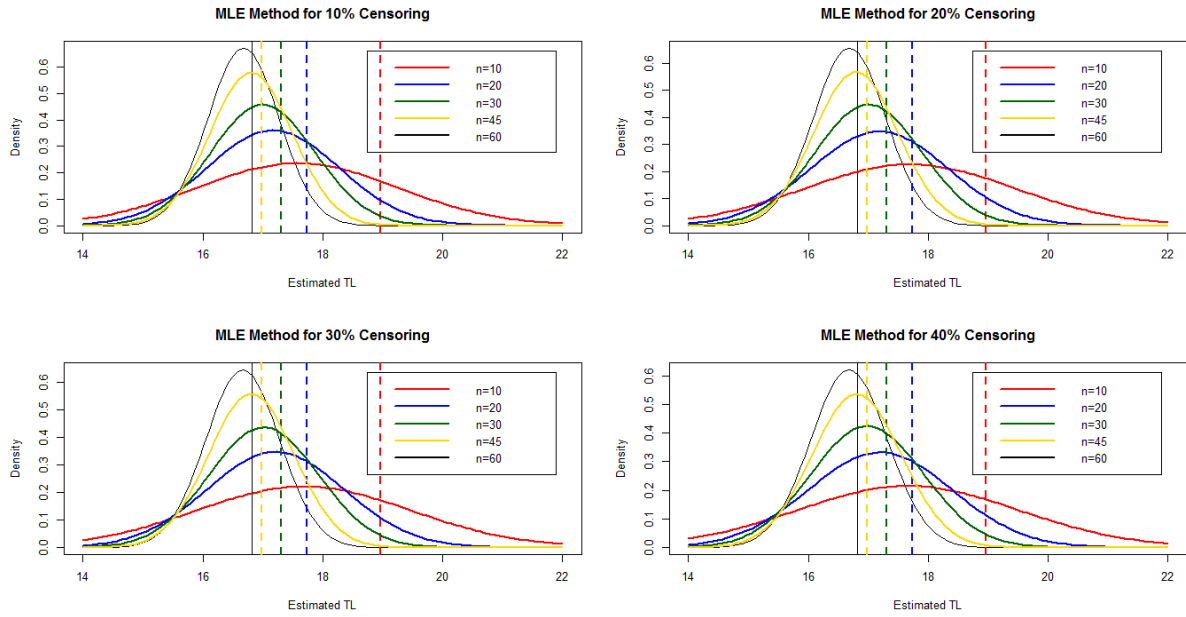


4.2 Evaluation of MLE Method

The MLE method has been believed to be the gold standard method in estimating population mean and variance. Its asymptotical unbiased property made the MLE method widely used in censored data analysis. The MLE method is almost unbiased (if there is any bias) in estimating the mean and deviates from the true population standard deviation by less than 10%. (See Appendix C) However, in estimating the UTL, the MLE method requires too much data to provide an acceptable coverage rate. It tends to underestimate the UTL and has a coverage rate with less than the 95% confidence level.

In Figure 4.2, the broken line (reference line) is the theoretical UTL calculated based on the true known population parameters. The curves represent the distribution of estimated UTLs. Since the center of every curve is on the left of its corresponding reference line, the MLE method is underestimating the UTL and hence has less than 95% cover rate of the 90th percentile. The figure does suggest a decreasing trend in both biasedness and standard error of the MLE as sample size increases.

Figure 4.2 Distribution of UTL estimated by MLE method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.



4.3 Evaluation of Traditional Substitution Methods

The traditional substitution methods have been wildly criticized for their biasedness in estimating the mean. It is not surprising to see that substituting censored observations by $\frac{1}{2}$ LOQ underestimates the mean and the LOQ method overestimates the mean. (See Appendix C) Hence, it is intuitive to believe that the $\frac{1}{2}$ LOQ substitutions will be liberal by underestimating the UTL and give a less than 95% coverage rate. Similarly, the LOQ substitutions will give conservative results. We have already shown the simulation results are the opposite of our previous expectations in terms of coverage rate.

A possible explanation of this counter-intuitive result is the bias in estimating the population variance. Table 4.2 suggests that the $\frac{1}{2}$ LOQ substitution underestimates the mean but overestimates the standard deviation; the LOQ substitution overestimates the mean and underestimates the standard deviation. Since the calculation of the UTL involves estimating both population mean and standard deviation, it is hard to tell in which direction a traditional substitution will be biased.

Unlike the MLE method, increasing sample size does not reduce biasedness for the traditional substitution methods. On the contrary, it tends to increase the biasedness as sample size increases. The distances between the reference line and the sample distribution of UTLs are increasing as sample size increases. (See Figure 4.3 and Figure 4.4)

Figure 4.3 Distribution of UTL estimated by $\frac{1}{2}$ LOQ substitution method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.

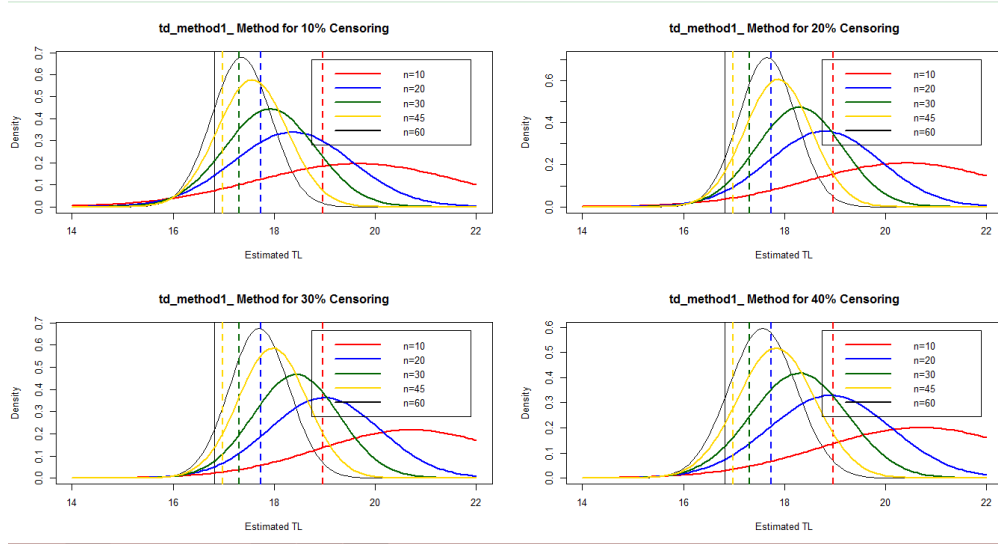
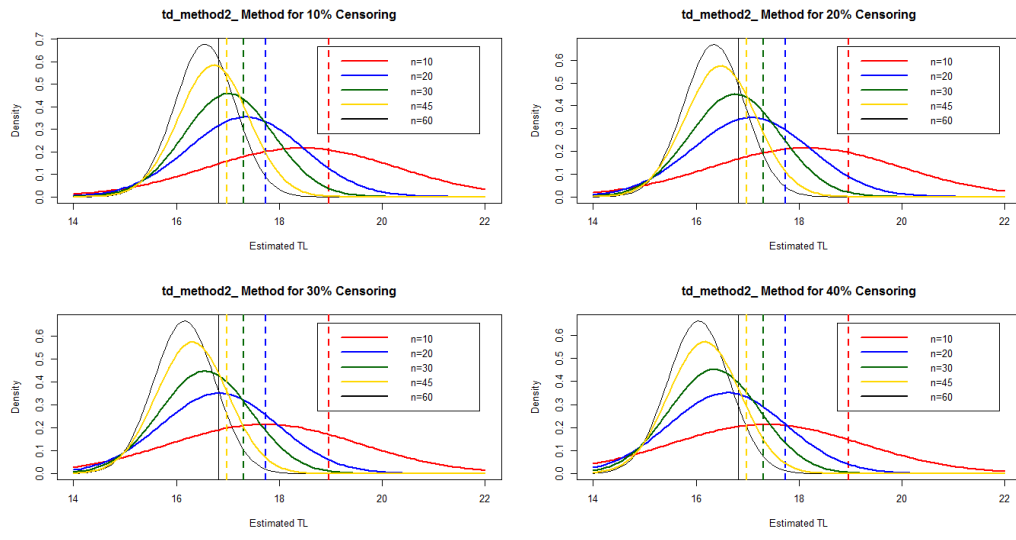


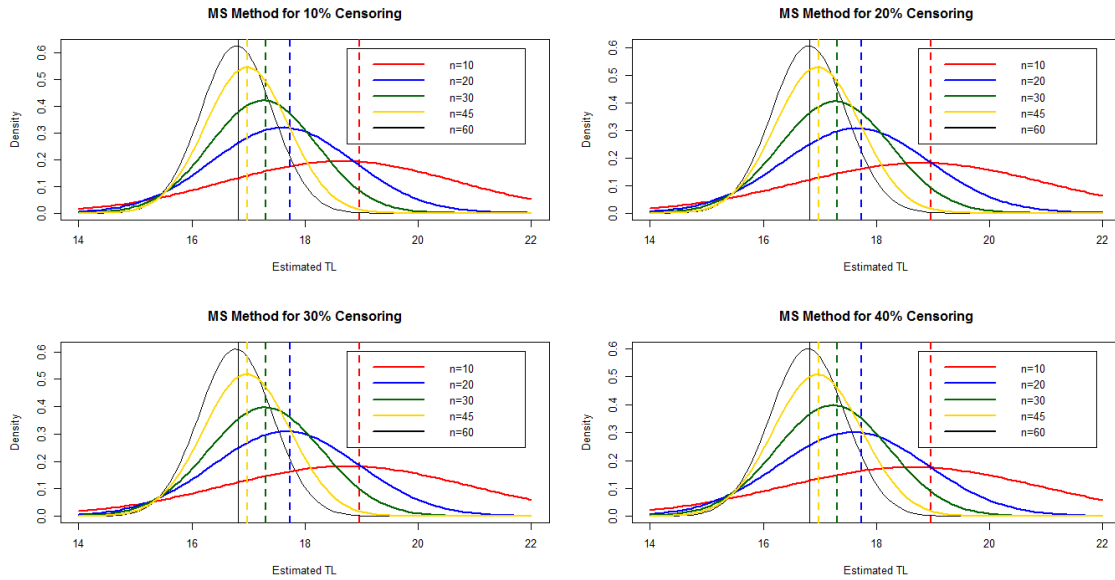
Figure 4.4 Distribution of UTL estimated by LOQ substitution method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$). Vertical lines are UTLs calculated using true population parameters.



4.4 Evaluation of Modified Substitution Method

The MS method we proposed in this report is slightly biased in estimating the mean. In table 4.3, most sample means are greater than $\mu = 12$ but not by much which suggests a small amount of overestimation. The sample standard deviation quickly approaches $\sigma = 3$ from the left as sample size increases. Figure 4.5 suggests that the percent of censored data does not have a strong effect on the MS method and its biasedness in estimating the UTL. The sample distribution of the UTL is always centered close to the reference line. The standard error of the UTL decreases as sample size increases which suggests a more reliable estimation of the UTL.

Figure 4.5 Distribution of UTL estimated by modified substitution method in different scenarios (percent censoring = 10%, 20%, 30%, 40%, $n = 10, 20, 30, 45, 60$).



4.5 Conclusions

Based on the simulation results, we have the following conclusions.

1. The MLE method is not appropriate for estimating the UTL when the sample size is small. The bias in the estimation of the mean and standard deviation is not too large for sample sizes of 20 to 45, but the bias of the UTL is too large to provide satisfactory coverage rate of the target percentile. The MLE method tends to underestimate the true UTL and provides a confidence level less than desired.
2. The traditional substitution methods are severely biased in estimating tolerance limits and the direction of biasedness is not clear. Even large sample sizes do not correct the bias. Therefore, the traditional substitution methods are not recommended.
3. The modified substitution method is less biased in estimation of the mean and standard deviation. It also provides a good coverage rate of the target percentile even when sample size is small. However, there are limitations. In order to provide substitutes for the censored observations, we need to be able to properly estimate the sample median which requires less than 50% of the data to be censored. Also, the modified substitution method requires the target population to be symmetric or can be transformable into a symmetric distribution.

Chapter 5 - Case Study and Discussion

Left censoring occurs not only in studies of food-production animals. The methods discussed in this report can be applied into other fields of study involving modeling low-concentration chemicals.

For example, table 5.1 contains part of the data collected from an environmental quality study given in Journal of Environmental Quality (Junk et al. 1980). Atrazine concentrations in a series of Nebraska wells before and after the growing season were measured. *Censor* = 1 indicates the observation is censored and *Censor* = 0 indicates it is the actual observed value. Suppose we want to estimate UTL to determine whether or not the Atrazine concentration is below the safety level at a particular month, e.g June.

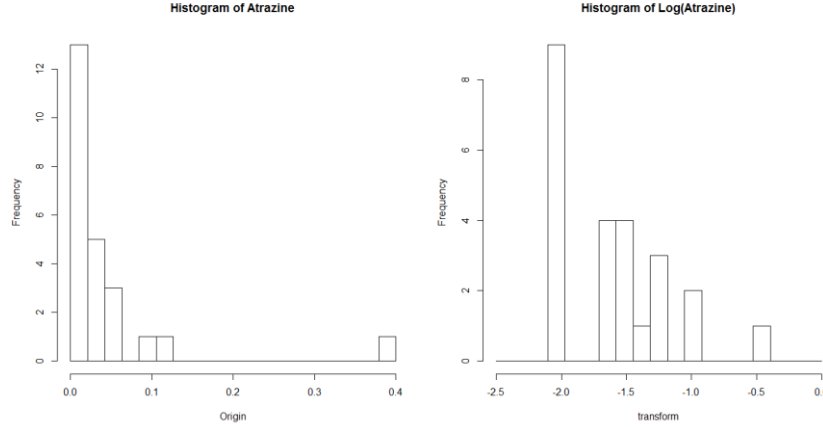
Table 5.1 Sample atrazine concentrations in Nebraska wells collected in June. Censor = 1 if the observation is censored and Censor = 0 indicates it is the actual observed value.

Atrazine	Month	Censor	Atrazine	Month	Censor
0.38	June	0	0.01	June	1
0.04	June	0	0.01	June	1
0.01	June	1	0.01	June	1
0.03	June	0	0.01	June	1
0.03	June	0	0.02	June	0
0.05	June	0	0.03	June	0
0.02	June	0	0.02	June	0
0.01	June	1	0.02	June	0
0.01	June	1	0.05	June	0
0.01	June	1	0.03	June	0
0.11	June	0	0.05	June	0
0.09	June	0	0.01	June	1

The LOQ in this study is 0.01 and there are 9 out of 24 (37.5%) observations that can be considered to be left censored. As shown in Figure 5.1, atrazine is not normally distributed, but the data after log transformation looks approximately normal. The sample mean, standard deviation, 95th percentile and (95, 95) UTL are calculated by using the MLE method and the modified substitution method. According to MLE method, $\bar{x} = -1.75$, $s = 0.60$, $\hat{p}_{95} = -0.78$,

UTL = -0.40 . By the modified substitution method, we have $\bar{x} = -1.69$, $s = 0.53$, $\hat{p}_{95} = -0.81$, UTL = -0.46. Hence, we are 95% confident that 95% of the wells have atrazine concentrations in June below $10^{UTL} = 0.40$ (by MLE method) $10^{UTL} = 0.45$ by modified substitution method.

Figure 5.1 Distribution of atrazine and log(Atrazine) for data in Table 5.1



In this report, we have examined several methods of dealing with left censored data in the estimation of an upper tolerance limit. We have shown that the modified substitution method may be preferred for a symmetric distribution where censoring does not exceed 50%. There are several suggestions for future study. This report does not provide theoretical justifications for the modified substitution method. However, it is possible that the modified substitution method is unbiased in estimating mean and population mean and asymptotically unbiased in estimating population variance as well as tolerance limit. The MLE method evaluated in this report is based on its asymptotic properties of MLEs. Another possible MLE method is to use the MLE invariance property to calculate the UTL directly. The Maximum Likelihood Estimators $\hat{\mu}$ and $\hat{\sigma}^2$ are μ and σ^2 that maximize the likelihood function $L(\mu, \sigma)$. As introduced in Chapter 1, the MLE of the p^{th} percentile is $\hat{P}_p = \bar{x} + Z_p \cdot s$, where Z_p is the p^{th} percentile of standard normal distribution $N(0,1)$; Because for any MLE estimator $\hat{\theta}$, the MLE estimator of $f(\theta)$ is $f(\hat{\theta})$ for any real function $f(\cdot)$ (Casella & Berger 2002), the MLE estimator of the UTL can be calculated as $UTL = \bar{x} + k_1 s$, where k_1 is defined as before. The modified substitution method

introduced in this report is a naive way to replace censored observations but appears to perform well compared to methods in current use.

Bibliography

- Casella, G., & Berger, R. L. (2002). Statistical inference (2nd ed.). Australia: Thomson Learning.
- CLSI. (2004). *Protocols for Determination of Limits of Detection and Limits of Quantitation, Approved Guideline*. Wayne, PA USA: Clinical and Laboratory Standards Institute.
- Hewett, P., & Ganser, G. (2007). A Comparison of Several Methods for Analyzing Censored Data. *British Occupational Hygiene Society*, 51(7), 611-632.
- Junk, G.A., Spalding, R.F., & Richard, J.J. (1980). Areal, vertical, and temporal differences in ground-water chemistry. *Journal of Environmental Quality*, 9, p, 479-483.
- Krishnamoorthy, K., & Lian, X. (2011). Closed-form approximate tolerance intervals for some general linear models and comparison studies. *Journal of Statistical Computation and Simulation*, 82(4), 547-563.
- Krishnamoorthy, K., & Mathew, T. (2009). *Statistical tolerance regions: theory, applications, and computation*. Hoboken, N.J.: Wiley.
- Lambert, D., Peterson, B., & Terpenning, I. (1991). Nondetects, Detection Limits, and the Probability of Detection. *Journal of the American Statistical Association*, 86, 414.
- WHO., & FAO. (2009). Maximum Residue Limits For Pesticides and Veterinary Drugs. *Principles and Methods for the Risk Assessment of Chemicals in Food* (pp. 8-2). WHO: Food and Agriculture Organization of the United Nations and World Health Organization.
- Wolynetz, M.S. (1979) Maximum Likelihood estimation in a Linear model from Confined and Censored Normal Data. *J.Roy.Statist.Soc (Series C)*, 28(2), 195–206

Appendix A - Summary of Simulation Results

The following tables summarized the simulation results. Recall that we have run the simulation 2500 rounds for each combination of percent of censoring and sample size. The hypothetical population from which random samples are drawn is $N(12, 3)$.

Table A.1 Mean of \bar{x} , estimated bias of \bar{x} , mean s, estimated bias of s, mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 10$.

$n = 10$								
LOQ=8.1553 (10% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.98	0.13%	2.80	6.62%	17.57	7.32%	84.36%
	MS	12.09	0.71%	2.78	7.24%	18.64	1.71%	92.32%
	$\frac{1}{2}$ LOQ	11.76	2.01%	3.35	11.73%	19.65	3.63%	96.76%
	LOQ	12.15	1.27%	2.67	11.02%	18.44	2.77%	92.16%
LOQ=9.4751 (20% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.98	0.15%	2.85	4.92%	17.70	6.68%	85.48%
	MS	12.09	0.73%	2.85	5.03%	18.80	0.88%	92.28%
	$\frac{1}{2}$ LOQ	11.44	4.70%	3.81	26.94%	20.40	7.59%	99.20%
	LOQ	12.37	3.07%	2.45	18.36%	18.14	4.36%	89.04%
LOQ=10.4268 (30% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.90	0.87%	2.88	4.08%	17.68	6.76%	84.80%
	MS	12.07	0.58%	2.83	5.63%	18.74	1.20%	91.96%
	$\frac{1}{2}$ LOQ	10.98	8.51%	4.14	37.86%	20.72	9.26%	99.48%
	LOQ	12.56	4.70%	2.19	27.09%	17.71	6.59%	83.80%
LOQ=11.2400 (40% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.84	1.35%	2.89	3.74%	17.66	6.87%	84.00%
	MS	12.16	1.37%	2.75	8.49%	18.63	1.76%	89.60%
	$\frac{1}{2}$ LOQ	10.55	12.09%	4.30	43.48%	20.68	9.08%	98.36%
	LOQ	12.84	6.96%	1.92	35.95%	17.36	8.45%	78.72%

Table A.2 Mean of \bar{x} , estimated bias of \bar{x} , mean s, estimated bias of s, mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 20$.

n=20								
LOQ=8.1553 (10% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.96	0.30%	2.91	2.85%	17.17	3.17%	88.20%
	MS	12.02	0.16%	2.90	3.30%	17.61	0.70%	92.56%
	$\frac{1}{2}$ LOQ	11.71	2.42%	3.46	15.19%	18.37	3.58%	98.48%
	LOQ	12.12	1.03%	2.71	9.59%	17.35	2.16%	90.56%
LOQ=9.4751 (20% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.97	0.24%	2.91	2.98%	17.19	3.07%	87.72%
	MS	12.02	0.16%	2.92	2.59%	17.65	0.47%	91.88%
	$\frac{1}{2}$ LOQ	11.39	5.06%	3.84	28.05%	18.79	5.99%	99.80%
	LOQ	12.33	2.74%	2.46	17.90%	17.07	3.71%	85.84%
LOQ=10.4268 (30% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.96	0.37%	2.92	2.51%	17.21	2.94%	88.64%
	MS	12.01	0.09%	2.94	2.16%	17.66	0.37%	92.96%
	$\frac{1}{2}$ LOQ	11.00	8.35%	4.15	38.38%	18.99	7.12%	99.68%
	LOQ	12.57	4.71%	2.22	26.14%	16.83	5.06%	80.72%
LOQ=11.2400 (40% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.93	0.59%	2.93	2.43%	17.20	3.00%	86.84%
	MS	12.08	0.69%	2.86	4.72%	17.59	0.81%	90.84%
	$\frac{1}{2}$ LOQ	10.58	11.87%	4.33	44.20%	18.91	6.64%	99.20%
	LOQ	12.84	7.01%	1.95	34.84%	16.61	6.34%	74.40%

Table A.3 Mean of \bar{x} , estimated bias of \bar{x} , mean s, estimated bias of s, mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 30$.

n=30								
LOQ=8.1553 (10% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.01	0.06%	2.95	1.76%	17.00	1.76%	91.52%
	MS	12.05	0.41%	2.93	2.29%	17.26	0.24%	93.88%
	0.5LOQ	11.76	2.04%	3.47	15.56%	17.92	3.56%	99.20%
	1.0LOQ	12.16	1.33%	2.73	8.95%	17.01	1.66%	91.52%
LOQ=9.4751 (20% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.00	0.04%	2.94	1.88%	17.00	1.73%	91.00%
	MS	12.05	0.37%	2.94	1.97%	17.27	0.17%	93.32%
	½ LOQ	11.41	4.89%	3.87	29.03%	18.29	5.73%	99.92%
	LOQ	12.35	2.93%	2.48	17.37%	16.76	3.14%	85.44%
LOQ=10.4268 (30% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.00	0.02%	2.95	1.73%	17.01	1.66%	90.20%
	MS	12.03	0.24%	2.96	1.27%	17.29	0.05%	92.60%
	½ LOQ	11.04	8.03%	4.16	38.70%	18.43	6.53%	99.80%
	LOQ	12.58	4.87%	2.24	25.43%	16.56	4.29%	78.08%
LOQ=11.2400 (40% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.97	0.29%	2.94	1.94%	16.98	1.85%	89.44%
	MS	12.05	0.46%	2.91	3.00%	17.23	0.43%	92.16%
	½ LOQ	10.60	11.63%	4.32	44.11%	18.29	5.71%	98.84%
	LOQ	12.85	7.07%	1.97	34.25%	16.35	5.47%	71.48%

Table A.4 Mean of \bar{x} , estimated bias of \bar{x} , mean s, estimated bias of s, mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 45$.

n=45								
LOQ=8.1553 (10% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.01	0.04%	2.97	1.08%	16.81	1.07%	91.72%
	MS	12.03	0.27%	2.96	1.37%	16.97	0.09%	94.48%
	½ LOQ	11.75	2.11%	3.49	16.21%	17.57	3.40%	99.36%
	LOQ	12.15	1.28%	2.74	8.73%	16.72	1.56%	89.56%
LOQ=9.4751 (20% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.98	0.15%	2.97	1.10%	16.79	1.14%	90.76%
	MS	12.00	0.03%	2.97	0.91%	16.97	0.13%	93.20%
	½ LOQ	11.38	5.17%	3.89	29.67%	17.87	5.21%	99.96%
	LOQ	12.33	2.75%	2.48	17.17%	16.48	3.00%	81.88%
LOQ=10.4268 (30% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.99	0.10%	2.97	1.12%	16.81	1.06%	91.28%
	MS	12.01	0.11%	2.97	1.01%	16.97	0.11%	93.00%
	½ LOQ	11.01	8.23%	4.17	38.98%	17.97	5.79%	99.92%
	LOQ	12.58	4.79%	2.24	25.44%	16.31	4.00%	75.12%
LOQ=11.2400 (40% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.97	0.25%	2.97	0.94%	16.81	1.07%	90.48%
	MS	12.02	0.16%	2.96	1.36%	16.96	0.17%	92.84%
	½ LOQ	10.60	11.68%	4.33	44.49%	17.83	4.98%	98.68%
	LOQ	12.85	7.10%	1.99	33.81%	16.17	4.84%	68.40%

Table A.5 Mean of \bar{x} , estimated bias of \bar{x} , mean s, estimated bias of s, mean UTL, estimated bias of UTL and cover rate of UTL for different levels of LOQ when $n = 60$.

n=60								
LOQ=8.1553 (10% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.01	0.05%	2.97	1.04%	16.68	0.82%	91.92%
	MS	12.03	0.25%	2.96	1.39%	16.79	0.14%	93.08%
	½ LOQ	11.75	2.11%	3.48	16.12%	17.35	3.21%	99.72%
	LOQ	12.15	1.27%	2.73	8.86%	16.55	1.55%	88.40%
LOQ=9.4751 (20% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	12.00	0.01%	2.97	0.88%	16.68	0.76%	91.36%
	MS	12.02	0.14%	2.98	0.81%	16.80	0.05%	92.72%
	½ LOQ	11.40	5.03%	3.89	29.70%	17.66	5.02%	99.92%
	LOQ	12.34	2.83%	2.49	17.00%	16.35	2.77%	80.16%
LOQ=10.4268 (30% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.98	0.15%	2.96	1.18%	16.66	0.90%	91.48%
	MS	12.00	0.00%	2.97	1.08%	16.77	0.23%	93.00%
	½ LOQ	11.00	8.36%	4.17	38.92%	17.70	5.29%	99.92%
	LOQ	12.56	4.69%	2.23	25.59%	16.15	3.91%	69.40%
LOQ=11.2400 (40% censored)	Method	Mean \bar{x}	mean bias%	Mean s	s bias %	UTL	UTL bias%	Coverage
	MLE	11.97	0.27%	2.98	0.68%	16.68	0.80%	90.48%
	MS	12.00	0.01%	2.97	0.88%	16.79	0.16%	91.68%
	½ LOQ	10.59	11.79%	4.34	44.58%	17.56	4.47%	99.32%
	LOQ	12.85	7.05%	1.99	33.70%	16.05	4.56%	63.80%

Appendix B - R code For Figure 4.2-4.5

R Code for figure 4.2

```
library(sas7bdat)
#####Importing data from SAS
rm(list=ls())
n=c(10,20,30,45,60)
p=c(10,20,30,40)
v=c(18.9626,17.7302,17.3016,16.9874,16.8126)

##Plot for mle method

#importing data

for (j in 1:4){
  for(i in 1:5) {
    infile <- paste("C:/Users/donglin/Desktop/Research/Graph
R/data/sim",j,"/", "mle",n[i], ".sas7bdat",sep="")

    temp=read.sas7bdat(infile)
    assign(paste("mle.p",p[j], ".n",n[i],sep=""),temp)

  } #the end of i loop
} #the end of j loop

#####plotting the MLE method 10% Censoring
```

```

P=c("P10","P20","P30","P40")
for (j in 1:4)
{
  a1=paste("mle.p",j*10,".n10",sep="")
  a2=paste("mle.p",j*10,".n20",sep="")
  a3=paste("mle.p",j*10,".n30",sep="")
  a4=paste("mle.p",j*10,".n45",sep="")
  a5=paste("mle.p",j*10,".n60",sep="")
  assign(paste("P",j*10,sep=""),c(a1,a2,a3,a4,a5))
}
windows()
par(mfrow=c(2,2))
for (index in 1:4){
x=seq(14,22,length=100)
y=dnorm(x,mean=mean(get(get(P[index]))[5])$TL),sd=sd(get(get(P[index]))[5])$TL))
plot(x, y, type="l", lty=1, xlab="Estimated TL",
      ylab="Density", main=paste("MLE Method for ",index*10,"% Censoring",sep=""))
abline (v=v[5],col="black")
colors <- c("red", "blue", "darkgreen", "gold", "black")
labels <- c("n=10", "n=20", "n=30", "n=45", "n=60")
for (a in 1:4){
  lines(x,
dnorm(x,mean=mean(get(get(P[index]))[a])$TL),sd=sd(get(get(P[index]))[a])$TL)), lwd=2,
col=colors[a])
  abline (v=v[a],lwd=2, lty=2,col=colors[a])
}
legend("topright", inset=.05,
      labels, lwd=2, lty=c(1, 1, 1, 1, 1), col=colors)
}

```

R Code for figure 4.3

```
library(sas7bdat)
#####Importing data from SAS
rm(list=ls())
n=c(10,20,30,45,60)
p=c(10,20,30,40)
v=c(18.9626,17.7302,17.3016,16.9874,16.8126)
##Plot for td_method1_ method

#importing data

for (j in 1:4){
  for(i in 1:5) {
    infile <- paste("C:/Users/donglin/Desktop/Research/Graph
R/data/sim",j,"/", "td_method1_",n[i],".sas7bdat",sep="")

    temp=read.sas7bdat(infile)
    assign(paste("td_method1_.p",p[j],".n",n[i],sep=""),temp)

  } #the end of i loop
} #the end of j loop

#####plotting the td_method1_ method 10% Censoring
P=c("P10","P20","P30","P40")
for (j in 1:4)
{
  a1=paste("td_method1_.p",j*10,".n10",sep="")
```

```

a2=paste("td_method1_.p",j*10,".n20",sep="")
a3=paste("td_method1_.p",j*10,".n30",sep="")
a4=paste("td_method1_.p",j*10,".n45",sep="")
a5=paste("td_method1_.p",j*10,".n60",sep="")
assign(paste("P",j*10,sep=""),c(a1,a2,a3,a4,a5))
}

windows()
par(mfrow=c(2,2))
for (index in 1:4){

x=seq(14,22,length=100)
y=dnorm(x,mean=mean(get(get(P[index]))[5])$TL),sd=sd(get(get(P[index]))[5])$TL))

plot(x, y, type="l", lty=1, xlab="Estimated TL",
      ylab="Density", main=paste("td_method1_ Method for ",index*10,"%
Censoring",sep=""))
abline (v=v[5],col="black")
colors <- c("red", "blue", "darkgreen", "gold", "black")
labels <- c("n=10", "n=20", "n=30", "n=45", "n=60")

for (a in 1:4){
  lines(x,
dnorm(x,mean=mean(get(get(P[index]))[a])$TL),sd=sd(get(get(P[index]))[a])$TL)), lwd=2,
col=colors[a])
  abline (v=v[a],lwd=2,lty=2,col=colors[a])
}
legend("topright", inset=.05,
      labels, lwd=2, lty=c(1, 1, 1, 1, 1), col=colors)
}

```

R Code for figure 4.4

```
library(sas7bdat)
#####Importing data from SAS
rm(list=ls())
v=c(18.9626,17.7302,17.3016,16.9874,16.8126)
##Plot for td_method2_ method
#importing data
for (j in 1:4){
  for(i in 1:5) {
    infile <- paste("C:/Users/donglin/Desktop/Research/Graph
R/data/sim",j,"/", "td_method2_",n[i], ".sas7bdat",sep="")
    temp=read.sas7bdat(infile)
    assign(paste("td_method2_.p",p[j], ".n",n[i],sep=""),temp)
  } #the end of i loop
} #the end of j loop

#####plotting the td_method2_ method 10% Censoring
P=c("P10","P20","P30","P40")
for (j in 1:4)
{
  a1=paste("td_method2_.p",j*10,".n10",sep="")
  a2=paste("td_method2_.p",j*10,".n20",sep="")
  a3=paste("td_method2_.p",j*10,".n30",sep="")
  a4=paste("td_method2_.p",j*10,".n45",sep="")
  a5=paste("td_method2_.p",j*10,".n60",sep="")
  assign(paste("P",j*10,sep=""),c(a1,a2,a3,a4,a5))
}
windows()
par(mfrow=c(2,2))
for (index in 1:4){
```



```

x=seq(14,22,length=100)
y=dnorm(x,mean=mean(get(get(P[index]))[5])$TL),sd=sd(get(get(P[index]))[5])$TL))

plot(x, y, type="l", lty=1, xlab="Estimated TL",
      ylab="Density", main=paste("td_method2_ Method for ",index*10,"%
Censoring",sep=""))
abline (v=v[5],col="black")
colors <- c("red", "blue", "darkgreen", "gold", "black")
labels <- c("n=10", "n=20", "n=30", "n=45", "n=60")

for (a in 1:4){
  lines(x,
        dnorm(x,mean=mean(get(get(P[index]))[a])$TL),sd=sd(get(get(P[index]))[a])$TL), lwd=2,
        col=colors[a])
  abline (v=v[a],lwd=2,lty=2,col=colors[a])
}

legend("topright", inset=.05,
      labels, lwd=2, lty=c(1, 1, 1, 1, 1), col=colors)
}

```

R Code for figure 4.5

```
library(sas7bdat)
#####Importing data from SAS
rm(list=ls())
n=c(10,20,30,45,60)
p=c(10,20,30,40)
v=c(18.9626,17.7302,17.3016,16.9874,16.8126)
##Plot for ms method

#importing data

for (j in 1:4){
  for(i in 1:5) {
    infile <- paste("C:/Users/donglin/Desktop/Research/Graph
R/data/sim",j,"/", "ms",n[i], ".sas7bdat",sep="")

    temp=read.sas7bdat(infile)
    assign(paste("ms.p",p[j], ".n",n[i],sep=""),temp)

  } #the end of i loop
} #the end of j loop

#####plotting the ms method 10% Censoring
P=c("P10","P20","P30","P40")
for (j in 1:4)
{
  a1=paste("ms.p",j*10,".n10",sep="")
```

```

a2=paste("ms.p",j*10,".n20",sep="")
a3=paste("ms.p",j*10,".n30",sep="")
a4=paste("ms.p",j*10,".n45",sep="")
a5=paste("ms.p",j*10,".n60",sep="")
assign(paste("P",j*10,sep=""),c(a1,a2,a3,a4,a5))
}

windows()
par(mfrow=c(2,2))
for (index in 1:4){

x=seq(14,22,length=100)
y=dnorm(x,mean=mean(get(get(P[index]))[5])$TL),sd=sd(get(get(P[index]))[5])$TL))

plot(x, y, type="l", lty=1, xlab="Estimated TL",
      ylab="Density", main=paste("MS Method for ",index*10,"% Censoring",sep=""))
abline (v=v[5],col="black")
colors <- c("red", "blue", "darkgreen", "gold", "black")
labels <- c("n=10", "n=20", "n=30", "n=45", "n=60")

for (a in 1:4){
  lines(x,
dnorm(x,mean=mean(get(get(P[index]))[a])$TL),sd=sd(get(get(P[index]))[a])$TL)), lwd=2,
col=colors[a])
  abline (v=v[a],lwd=2,lty=2,col=colors[a])
}

legend("topright", inset=.05,
      labels, lwd=2, lty=c(1, 1, 1, 1, 1), col=colors)
}

```

Appendix C - Simulation SAS code

```

%let content=0.9; %let K=2.21 ;**K value is calculated using
(1/sqrt(12))*qt(0.95,df=11,ncp=2*qnrm(0.90,0,1)*sqrt(12));
%let pop_mean=12;%let pop_sd=3;

options nonotes; /* prevents the SAS log from overflowing */

%macro Simulate(N, Rounds,seed,LOD);**temperaly set censoring limit to be 10;
%do j = 1 %to &Rounds;
  /* repeat many times */
  data temp&j;
    call streaminit(&seed+&j);
  /* 1. create a sample of size &N */
  do i =1 to &N;
    x=rand("normal",&pop_mean,&pop_sd);** mu+sigma*Z;

    upper=x;lower=x;
    if x <=&LOD then upper=&LOD;
    if x<=&LOD then lower=.;round=&j;
  output;
  end;run;

  /*****THE MLE
Method*****/
  ** result stored in data set MLE;
  proc lifereg data=temp&j outest=T;**T contains the MLE estimation of mean and
standard deviation;
    model (lower,upper)=/dist=normal;
    output out=O1 Q= &content P=Predi STD=stdd; **Output Estimated TL into data set
O1;
  run;

```

```

data T2; set T; Mean=Intercept; S=_Scale_; keep Mean S; run;
data O2; **O2 contains the statistic from MLE;
set O1;
where i=1;
TL=predi+1.645*stdd; keep TL _PROB_ Predi stdd; run;
proc append base=X1 data=o2; run;
proc append base=X2 data=T2; run;
data MLE&N; merge X1 X2; run;
** use PROC APPEND to accumulate statistics ;

/*****The End of MLE
Method*****/

/***** the MS
method*****/

proc summary data=temp&j median;
var upper;
output out=medians (drop=_) median=med;
run;
proc sql;
create table a as
select upper as ac
from Temp&j
order by ac asc;
create table b as
select upper as dc
from temp&j
order by dc desc;
create table C as
select ac, med from A, Medians;
quit ;

```

```

data MSsubst;
merge C B;
if ac=&LOD then sub=2*med-dc;
else if ac>&LOD then sub=ac;
keep sub med;
run;
*ods trace on;*ods trace off;
ods output quantiles=Q;
proc univariate data=MSsubst ciquantnormal(type=upper alpha=0.05);
var sub;
output out=MS2 mean=mean std=s;
run;
ods output close;
data MS1;
set Q;
where Quantile="90% ";
predi=estimate; TL=UCLnormal;
keep varname predi TL;
run;
data MS3;
merge MS1 MS2;
run;
proc append base=MS&N data=MS3;run;

*****The end of MS method*****;
/***** the Traditional Substisution
method*****/

data traditional_sub;

```

```

set temp&j;
if lower=. then do; Y1=(1/2)*&Lod; Y2=&Lod;end;
else if lower ne . then do; Y1=lower; Y2=lower;end;
output;run;

ods output quantiles=trad_Q1;
proc univariate data=traditional_sub ciquantnormal(type=upper alpha=0.05);
var Y1;
output out=TD1_1 mean=mean std=s ;
run;
ods output close;
data TD1_2;
set trad_Q1;
where Quantile="90%";
predi=estimate; TL=UCLnormal;
keep varname predi TL;
run;
data TD_1_3;
merge TD1_1 TD1_2;
run;
proc append base=TD_METHOD1_&N data=TD_1_3;run;

ods output quantiles=trad_Q2;
proc univariate data=traditional_sub ciquantnormal(type=upper alpha=0.05);
var Y2;
output out=TD2_1 mean=mean std=s ;
run;
ods output close;
data TD2_2;
set trad_Q2;
where Quantile="90%";

```

```

predi=estimate; TL=UCLnormal;
keep varname predi TL;
run;
data TD_2_3;
merge TD2_1 TD2_2;
run;
proc append base=TD_METHOD2_&N data=TD_2_3;run;

*****the end of traditional Substitution
Methods*****;

*****Control Group- No censored
data*****;
*****No censored data*****;

ods output quantiles=trad_Q0;
proc univariate data=traditional_sub ciquantnormal(type=upper alpha=0.05);
var X;
output out=TD0_1 mean=mean std=s ;
run;
ods output close;
data TD0_2;
set trad_Q0;
where Quantile="90% ";
predi=estimate; TL=UCLnormal;
keep varname predi TL;
run;
data TD_0_3;
merge TD0_1 TD0_2;

```



```

run;

proc append base=TD_METHOD0_&N data=TD_0_3;run;

*****The end of no censored
Data*****;
*****
*****;

proc datasets nolist;
delete Temp&j;
%end;

proc datasets nolist;
delete O1 O2 X1 X2 T T2 TD1_1 TD1_2 TD_1_3 TD2_1 TD2_2 TD_2_3 TraD_q1
TraD_q2 traditional_sub/*from MLE*/ MS1 MS2 MS3 Q Medians A B C MSsubst /*from MS*/
TD0_1 TD0_2 TD_0_3 TRAD_Q0;
run;

options notes;/* re-open notes */
%mend;

%macro auto(); **
%let n1=10;%let n2=20;%let n3=30; %let n4=45; %let n5=60;
%let seed1=600874;
%let seed2=220794;
%let seed3=19891014;
%let seed4=2849172;*;
%let seed5=8463840;*;
%do indx=1 %to 5;
%let n=n&indx; %let seed=seed&indx;
%Simulate(&&&n,2,&&&seed,LOD=10.4268);run; **N, ROUND, SEED, LOD (sd);
%end;
%mend auto;

%auto();

```

```

%macro EV(Sigma,Percent,lib);
%let n1=10;%let n2=20;%let n3=30;?%let n4=45; %let n5=60;

%do indx=1 %to 5;
%let N=n&indx;
title "Evaluation of MLE method (Sigma=&sigma, Percent=&Percent%)" ;
title2 "sample size =&&&n";

data MLE_E&&&N;
set sim.mle&&&N; ** resulting data are stored in library = result;
if tl>=12+3*quantile('normal',0.9) then cover=1;
else if tl<12+3*quantile('normal',0.9) then cover=0;
run;
title "Evaluation of MLE method (Sigma=&sigma, Percent=&Percent%)" ;
title2 "sample size =&&&n";
proc means data=MLE_e&&&N ;
var MEAN S tl cover;
output out=temp1;
run;
data temp2;
set temp1 (keep= mean s tl cover);
if _n_ ne 4 then delete;
Sigma=&sigma; Percent=&percent; n=&&&n; method="MLE";
run;

proc append base=MLE_summary data=temp2;run;
%end;
%mend EV;
%EV(Sigma=3,Percent=10);

```